Objection to Specification

The Examiner has objected to the specification under §112, first paragraph, as failing to provide an adequate written description to enable the present invention. The applicants respectfully disagree with this assertion.

The basis of the Examiner's rejection is the fact that the applicants recited in Table I of the specification their results from the procedures used to make transgenic plants, and it is listed that toxicity was not obtained with all plants. The Examiner asserts from this fact that the applicants specification is not enabling, since not all plants demonstrated toxicity to insects. The applicants respectfully disagree. The specification is fully enabling and the results set forth in Table I are not differ in character from analogous results obtained by all others of skill in the art.

It is a fact well known to those of ordinary skill in the art that the yield of plants from a plant transformation procedure will include plant lines which vary over a range of level of expression of the desired protein or gene product. Such processes will also normally yield some percentage of poorly expressing or non-expressing plants. The reasons for this variation are not all clear, but some of the reasons include variation in inserted copy number and positional effects from adjacent indigenous plant genes on the inserted genes. This effect is inherent in technologies which, like all the presently available technologies for plant genetic engineering, rely on random location non-specific insertion of the gene constructs into the plant genome. While each individual plant line produced from each transgenic plant is stable as to its level of expression, the level of expression from plant line to plant line will vary. Each first generation transgenic plant can, by self-pollination and selection of progeny, be made to give rise to such a line.

Note that the applicants' results are not different in this respect from those of the prior art. The Examiner has cited the paper by Veack, et al., which contains on page 35 a series of bar graphs which demonstrate that several plants were relatively toxic, while others were insignificantly toxic compared to the controls.

Note also that the results listed on Table I are a reflection of the stringency of the applicants' assay method. The applicants used only larvae which had been previously matured by feeding on non transgenic

8.1

plants, since neo-natal larvae are more sensitive (specification page 18), in contrast to Veack et al., who used freshly hatched (i.e. more sensitive) larvae for their assay.

In any event, the reported processes and results do not fail to enable the present invention. Although not all transgenic plants which survived selection on a selection agent expressed the toxin at desired levels, clearly sufficient number of the plants did express the toxin at significant level so as to demonstrate that fully expressing plants can be readily achieved without the need for undo experimentation. It is not and has never been a requirement of section 112 that a patentable process must work in every instance, every time, without fail. Instead, the standard is that the process must be enable so that one of ordinary skill in the art to recreate the process without undue experimentation, and the present specification is sufficiently detailed to pass that test. Accordingly the rejection under §112 is believed not well founded.

Rejection under §103.

The Office Action contains a statement regarding the assumption of commonly owned subject matter. All the subject matter of this patent application has at all times been commonly owned.

The Examiner rejected all the claims pending in this patent application based on a combination of five cited publications. The first publication, to Hoekma et al, is cited to illustrate the concept of codon preference in yeast. The paper to Schnepf et al. is cited insofar as it shows that the Bacillus thuringiensis delta-endotoxin coding region is rich in A and T nucleotides. Murray is cited to show that plant genes are known to be rich in G and C nucleotides. Murray is cited to suggest modification of nucleotide structure to increase expression.

There are several flaws with the rejection as applied by the Examiner. One significant flaw is that the Murray et al. paper is not properly a reference against the claims of the present patent application. Since the paper was not published more than one year prior to the filing date of this application, presumably the paper was cited in the event the paper might be a reference under 35 U.S.C. §102(a). Submitted herewith is a Declaration of Michael J. Miller, one of the inventors of the present subject matter, with sufficient evidence under Rule §1.131 so as to make it

clear that the Murray et al. paper is not properly a reference against the claims of the present patent application. Accordingly any rejection which relies on this paper is improper.

The Examiner's principle reference was the paper by Hoekema et al. That paper discloses that a highly expressed gene in the yeast Saccharomyces cerevisiae could be have its level of expression reduced if the codon usage pattern of the gene was altered. The applicants assert that this reference alone is insufficient to make obvious the method and the plants of the present invention, and that the other properly cited references do not suffice to overcome this shortcoming.

The Examiner cites the Vaeck at al. reference for the proposition that the Bacillus thuringiensis delta endotoxin is difficult to express well in plants. The applicants admit that this is the case. However, the pattern of codon usage is only one of the mechanisms to which this poor expression level may be ascribed. It must be remembered that this toxin is, in fact, a toxin. In the applicants' experience, no transgenic plant could be created at all which expressed the full-length B.t. protoxin, even though transgenic plants can readily be recovered which express the amino terminal portion of the protoxin, referred to as the toxin segment. One very logical explanation for this result is toxicity to plant cells of the full length protoxin. If the protein is toxic at any level to plant cells, that could certainly be a factor in poor expression in transgenic plants, since highly expressing cells might simply either be dying or be selected against in regeneration. Other factors, such as poor mRNA stability, or poor protein stability could also explain the poor expression of the B.t. gene in plants. Accordingly, the teaching of Vaeck et al. does not make it clear that changes in the pattern of codon usage could cure the difficulties in expression of this gene in plants, which the applicants here have discovered to be true.

This analysis of the secondary Vaeck et al. reference help to demonstrate some of the inadequacies in the Examiner's argument that the Hoekma et al. makes obvious what the Examiner asserts it makes obvious. Hoekma et al. teaches that a highly expressed gene can be made to express poorly by substitution of codon usage, but that observation does not demonstrate that a poorly expressed gene could be made to express better by the reverse strategy, particularly when there are several other equally

plausible explanations for the poor expression. Secondly, Hoekma et al. discloses only the codon usage for one gene. This does not suggest that codon usage over a species, or over plants as a whole, is possible or would be effective. Note that the pattern of codon usage of the yeast gene of Hoekma is very different from the plant gene codon usage of the applicants' Fig. 1. Compare the pattern of codon usage for serine (plants prefer AGC, the yeast gene prefers TCT), glutamine (plants CAG, yeast CAA), glutamic acid (plants GAG, yeast GAA), arginine (plants CGC or AGG, yeast AGA), or glycine (plants GGC, yeast GGT). There are thus significant differences among organisms as diverse as yeast and plants in the pattern of codon usage. Does Hoekma teach then that the pattern of gene expression in each individual plant must be examined to develop effective data on codon usage? That is not what the applicants did. The applicants have analyzed the pattern of codon usage in native plants genes as a whole, not from any one species, and from that data, stretching throughout the plant kingdom, constructed a table of codon usage. The applicants then used that data to make a gene based on that pattern, and found it to be effective. Hoekma does not provide evidence from which one could predict that this would be possible. Hoekma does not teach that substituting codon usage would increase expression of poorly expressed genes, and does not suggest that a codon usage table incorporating data from plants stretching across species could be effective in increasing expression of a poorly expressed gene. The Examiner cites In re O'Farrell, 7 USPQ 2d 1673 (Fed. Cir. 1988) for the proposition that absolute predictability is not required, which is true, but what is required is more than prior art which gives no indication of which parameters are critical or which of many possible choices is likely to be successful, In re O'Farrell at 1681. The Hoekma reference does not provide sufficient information from which a reasonable expectation of success could be predicted. Accordingly this reference fails to make a case of prima facie obviousness for the claims of the present invention.

The applicants' assertion of non-obviousness applies with even more force to the specific constructions claimed in claims 15 and the new claim 17. In both those claims, the codon usage alteration of the gene is not recited as complete, but includes the amino terminal portion of the B.t. gene. Nothing in Hoekma, or in Vaeck, or in any of the other references, suggests that by changing the pattern of codon usage only at the amino

terminal portion of a difficult to express gene, such as the B.t. toxin, that significant increases in levels of expression could be achieved. This result is entirely unanticipated by the prior art, even if it were taken to suggest what the Examiner asserts it suggests. Accordingly, these claims are not in any way made obvious by the cited references.

Conclusion

Wherefore, again the Examiner is respectfully requested to revisit the merits of the specification and claims of this patent application. An early and favorable reply is solicited. A separate request for extension of time is submitted herewith so that this Amendment may be considered as timely filed.

Respectfully submitted,

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